

Branhamella (*Neisseria*) *catarrhalis*—a Lower Respiratory Tract Pathogen?

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Branhamella (*Neisseria*) *catarrhalis* was identified as a probable respiratory tract pathogen in seven patients, four with pneumonia and three with bronchitis. Five of the *B. catarrhalis* isolates produced β -lactamase. Production of β -lactamase correlated with penicillin resistance by the standardized disk diffusion method and also with high minimal inhibitory concentrations of penicillin for the two strains which were tested.

In the past year, we have observed two characteristics of *Branhamella* (*Neisseria*) *catarrhalis* which have not been widely recognized. The first is the apparent capacity to produce acute respiratory infections in patients with chronic lung disease, and the second is the ability to produce β -lactamase. In seven patients—all of whom had underlying pulmonary pathology—bronchitis or pneumonia was documented by clinical presentation, roentgenographic findings, and sputum examination. We used one method for plasmid extraction and found no plasmid deoxyribonucleic acid in the β -lactamase-positive or -negative strains of *B. catarrhalis*.

CASE REPORTS

Patient 1. Patient 1 was a 53-year-old man admitted 19 April 1978 with fever and an acute episode of wheezing following aspiration. In the past, the patient had undergone surgery for a giant hiatal hernia and now had an intrathoracic stomach. Recurrent episodes of aspiration pneumonia had prompted numerous hospitalizations. Examination upon this admission showed an acutely ill man in respiratory distress. Wheezing was heard throughout the chest. A chest X ray showed a large infiltrate in the right lower lobe and a smaller infiltrate in the left lower lobe. The patient was treated with bronchodilators, steroids, and penicillin. The sputum Gram stain showed 3+ polymorphonuclear leukocytes, 3+ gram-positive cocci in pairs and short chains, and 3+ gram-negative cocci (see below). The culture subsequently grew 3+ *Streptococcus pneumoniae* and 2+ oral flora. The patient improved only slightly after treatment with penicillin and then ampicillin, and he continued to aspirate. Repeated sputum Gram stains showed persistence of leukocytes and gram-negative cocci. Repeated cultures

grew only 3+ *B. catarrhalis* and were β -lactamase positive. Because of persistent fever, leukocytosis, and pulmonary infiltrates, therapy was changed to oral tetracycline to treat the *B. catarrhalis* infection. The subsequent course of the patient was one of gradual clinical and roentgenographic improvement, and he was discharged afebrile with a clear chest X ray after 7 days of oral tetracycline.

Patient 3. Patient 3 was a 75-year-old woman admitted 19 November 1978 with acute pulmonary edema. She had a history of diabetes mellitus, refractory congestive heart failure, cranial arteritis treated with steroids, and chronic bronchitis with emphysema. The patient responded to therapy for her congestive failure but was noted to have a productive cough 1 day after admission. She had no fever and no roentgenographic evidence of pneumonia. A sputum Gram stain revealed 4+ polymorphonuclear leukocytes and 4+ gram-negative diplococci. The patient was started on oral tetracycline, and there was an improvement of her cough and a decrease in sputum production. The sputum culture subsequently grew 3+ *B. catarrhalis* and was β -lactamase positive but susceptible to tetracycline.

MATERIALS AND METHODS

Patients with suspected *B. catarrhalis* infection were identified by reviewing sputum Gram stains and culture results. Charts of patients with apparent infection were then reviewed to obtain information on clinical course, roentgenographic findings, treatment, and outcome.

All sputum specimens received in the laboratory were initially processed in the same way. A purulent portion of sputum was selected for Gram staining and culturing. Blood and chocolate agars were inoculated and incubated in 5% CO₂. Another blood agar plate was incubated anaerobically, and a MacConkey agar plate was incubated aerobically. Quantitation of organisms, leukocytes, and epithelial cells by the Gram stain was assessed on a 1+ to 4+ scale: 1+ indicates 1 per oil immersion field; 2+ indicates 1 to 5 per oil

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immersion field; 3+ indicates 5 to 30 per oil immersion field; 4+ indicates 30 or more per oil immersion field. Quantitation of bacterial growth was assessed on a 1+ to 4+ scale: 1+ indicates light growth in the primary streaking zone of the original plate; 2+ indicates heavy growth in the primary streaking zone only; 3+ indicates growth in the primary and secondary streaking zones only; and 4+ indicates growth in the primary, secondary, and tertiary streaking zones.

B. catarrhalis was identified by the following characteristics: gram-negative diplococci; oxidase positive; growth on chocolate and blood agars, at 36°C in air or 5% CO₂; *o*-nitrophenyl- β -D-galactopyranoside negative; citrate negative; and nitrate reduced. Carbohydrate fermentation was tested with the Minitek system (BBL Microbiology Systems, Cockeysville, Md.) and was negative for glucose, maltose, and lactose. Identification of the isolates as *B. catarrhalis* was confirmed by the San Francisco Department of Public Health.

β -Lactamase was detected by a modification of the chromogenic cephalosporin procedure (11). Penicillin susceptibility was tested by the standardized disk diffusion method. For two of the β -lactamase-producing strains, susceptibility to penicillin was also tested by a broth dilution procedure utilizing inocula of 10⁴ and 10⁶ colony-forming units per ml.

To screen for the presence of extrachromosomal deoxyribonucleic acid, *B. catarrhalis* was grown overnight in the presence of penicillin (1 μ g/ml). Ethanol-precipitated deoxyribonucleic acid was prepared in duplicate from cleared lysates and was electrophoresed in a 0.7% agarose gel as described by Meyers et al. (10).

RESULTS

B. catarrhalis was identified as a probable respiratory tract pathogen in seven patients. In all seven patients identified as infected, sputum plus Gram stains showed 3+ to 4+ polymorphonuclear leukocytes, few epithelial cells, and 3+ to 4+ gram-negative diplococci located intracellularly and extracellularly. Other bacteria were seen only in small numbers (rarely to 1+), and *B. catarrhalis* was the predominant organism recovered in culture (3+ to 4+ growth).

Of the seven isolates of *B. catarrhalis*, five were found to produce β -lactamase. Penicillin resistance, as determined by the standardized disk diffusion method, correlated in all instances with the production of β -lactamase. For the two β -lactamase-producing strains which were tested by the broth dilution procedure, the minimal inhibitory concentrations for penicillin were 25 and 12 μ g/ml, respectively, with an inoculum of 10⁴ colony-forming units per ml, and 100 and 25 μ g/ml, respectively, with an inoculum of 10⁶ colony-forming units per ml.

No plasmid deoxyribonucleic acid could be detected in either β -lactamase-positive or -negative strains grown in broth or on agar containing penicillin.

Clinical data. Four of the patients from whom *B. catarrhalis* was isolated had pneumonia documented by chest roentgenography. The remaining three had acute bronchitis, defined as an increase in cough with production of large amounts of purulent sputum. Clinical data on all the patients are presented in Table 1. Patients 1 and 3 are presented as illustrative cases.

DISCUSSION

The recognition of *B. catarrhalis* as a possible pathogen in our patients prompted a review of the literature. *B. catarrhalis* has been isolated from middle ear aspirates in a small percentage of children with otitis media (7). Sporadic cases of meningitis (3, 4), endocarditis (14), urethritis (8), and bacteremia with and without purpura fulminans (1, 5) have also been reported. To our knowledge, only one patient with *B. catarrhalis* pulmonary infection has been previously reported from the United States (9). This patient was immunosuppressed, and he died from overwhelming *B. catarrhalis* pneumonia.

The role of *B. catarrhalis* in pulmonary infections has been demonstrated best by Ninane and colleagues in Belgium (12). These investigators performed transtracheal aspiration on retired coal miners with acute bronchitis and chronic lung disease. *B. catarrhalis*, in pure culture, was the third most common isolate from these patients. (*Haemophilus influenzae* and *S. pneumoniae* were isolated more frequently.) *B. catarrhalis* was also isolated in combination with *H. influenzae* in one patient. Of particular interest is the fact that one patient had β -lactamase-producing *B. catarrhalis* with a minimal inhibitory concentration of ampicillin of 16 μ g/ml. This patient failed to respond to ampicillin, but he improved rapidly when treated with cefuroxime (minimal inhibitory concentration, 2 μ g/ml). Ninane et al. reported another anthracosilicotic coal miner with acute bronchitis who had β -lactamase-producing *B. catarrhalis* grown in pure culture from a transtracheal aspirate (13). This patient responded to therapy with amoxicillin combined with clavulanic acid, a β -lactamase inhibitor.

All seven of our patients whose sputum yielded *B. catarrhalis* as the predominant organism had some underlying lung pathology. Most of these patients had received ampicillin or penicillin intermittently for past pulmonary infections. The finding of several β -lactamase-producing *B. catarrhalis* strains in this patient population may be related to this prior antibiotic usage; we postulate that prior penicillin use may have allowed selection of a resistant population

TABLE 1. *Clinical data*

Patient	Age (yr)	Sex	Underlying disease	X-ray finding	Sputum culture	Clinical diagnosis	Therapy	Outcome
1	53	M	Recurrent aspiration pneumonia	Right and left lower lobe infiltrates	3+ <i>B. catarrhalis</i> , β -lactamase +	Pneumonia	Ampicillin Tetracycline	No change Improved
2	71	F	Chronic obstructive lung disease, hip fracture	No infiltrate	4+ <i>B. catarrhalis</i> , β -lactamase -	Bronchitis	Ampicillin	Improved
3	75	F	Hypertension, diabetes, chronic bronchitis, congestive heart failure	No infiltrate	3+ <i>B. catarrhalis</i> , β -lactamase +	Bronchitis	Tetracycline	Improved
4	81	M	Arteriosclerotic heart disease, chronic obstructive lung disease	Lingular infiltrate	3+ <i>B. catarrhalis</i> , β -lactamase +	Pneumonia	Penicillin	Improved
5	59	F	Severe mitral stenosis, congestive heart failure	No infiltrate	3+ <i>B. catarrhalis</i> , β -lactamase +	Bronchitis	Tetracycline	Improved
6	73	F	Metastatic adenocarcinoma with pulmonary involvement	Right middle upper and lower lobe infiltrates	4+ <i>B. catarrhalis</i> , β -lactamase +	Pneumonia (behind obstruction)	Gentamicin, carbenicillin	Persistent infiltrates
7	81	F	Arteriosclerotic heart disease, chronic obstructive pulmonary disease	Right lower lobe infiltrate	2+ <i>B. catarrhalis</i> , β -lactamase -	Pneumonia	Tetracycline	Improved

of organisms. Since the patients were compromised in terms of pulmonary function, these organisms were then able to express their pathogenic potential.

In Gram stains of respiratory secretions from several of our patients, *B. catarrhalis* was mistaken for *Neisseria meningitidis*, a well-known respiratory tract pathogen (6, 15), and therapy with penicillin was initiated. Of two patients with pneumonia presumably caused by β -lactamase-producing strains, one did respond to high-dose intravenous penicillin therapy; thus, the question is raised of etiology or the susceptibility of β -lactamase-producing strains to high-dose penicillin. The other patient with pneumonia required a change in therapy from ampicillin to tetracycline before clinical improvement occurred.

β -Lactamase production by our strains was apparently not plasmid mediated. Similar findings have been reported for β -lactamase-producing *B. catarrhalis* strains isolated in France and England (2, 16). However, we did not try variations of the extraction procedures that may be necessary for demonstrating plasmids in this particular organism.

Further studies of the etiological role of *B.*

catarrhalis and the clinical importance of its β -lactamase production will require a prospective evaluation of a large group of patients with acute respiratory infections. Isolation of the organism from normally sterile areas, such as pleural fluid and blood, will resolve the question of pathogenicity. However, our preliminary observations suggest that *B. catarrhalis* may be another organism that is capable of causing acute bronchitis or pneumonia in patients with underlying lung disease.

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